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CANCER INCIDENCE OTHER THAN GASTRIC CANCER IN PERNICIOUS ANEMIA: A SYSTEMATIC REVIEW WITH META-ANALYSIS

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INTRODUCTION

Pernicious anemia (PA) is the end-stage of autoimmune gastritis (AG), likely as a consequence of long-standing *H pylori* infection. The active infectious process is gradually replaced by an autoimmune disease terminating in a burned-out infection and the irreversible destruction of the gastric body mucosa (Toh et al, 1998; Bergman et al, 2001). The autoimmune origin of PA is further supported by the presence of parietal cell and/or intrinsic factor autoantibodies and the frequent association with other autoimmune disorders, such as autoimmune thyroid disease, type I diabetes, and vitiligo (Toh et al, 1998; De Block et al, 2008; Lahner et al, 2008).

PA is a macrocytic anemia due to cobalamin (vitamin B_{12}) deficiency, which, in turn, is the result of deficiency of intrinsic factor, a protein that binds avidly to dietary vitamin B_{12} and promotes its transport to the terminal ileum for absorption. The deficiency of intrinsic factor is a consequence of the presence of atrophic gastritis resulting in the destruction of the oxyntic mucosa, and, thus, the loss of parietal cells, which normally produce chlorhydric acid as well as intrinsic factor (Annibale et al, 2011; Babior et al, 1998). The term PA is sometimes used as synonym for cobalamin deficiency or for macrocytic anemia, but to avoid ambiguity, PA should be reserved for conditions resulting from impaired secretion of intrinsic factor and atrophy of oxyntic mucosa. However, differential diagnosis may sometimes be challenging due to the limit of available diagnostic tools.

Pernicious anemia: epidemiology and clinical presentation

The clinical presentation of PA is often insidious for various reasons. The onset and progression of PA are very slow. As a consequence, often patients are not aware of their symptoms related to anemia, because over time they had got used to them. In many such cases, the underlying disease may not be suspected until a complete red blood count has been performed. However, patients with PA may seek medical advice due to aspecific symptoms related to the presence of anemia *per se*, such as weakness, asthenia, decreased mental concentration,

headache, and, especially in elderly patients, cardiological symptoms such as palpitations and chest pain (Annibale et al, 2011; Babior et al, 1998). Less frequently, PA may present with neurological symptoms, such as paresthesias, unsteady gait, clumsiness and in some cases spasticity. Indeed, vitamin B_{12} deficiency may cause peripheral neuropathy and lesions in the posterior and lateral columns of the spinal cord (subacute combined degeneration) and in the cerebrum, and these lesions progress from demyelisation to axonal degeneration and eventual neuronal death. Moreover, there is a growing body of evidence on the relationship between cobalamin deficiency and dementia (10). The early recognition of these symptoms is very important, because the neurological lesions may not be reversed after replacement therapy with vitamin B_{12} and patients with PA generally respond favorably to supplemental B_{12} treatment, especially if PA is diagnosed early in the course of the disease (Werder, 2010).

Finally, the onset of PA may be observed in patients under medical treatment for other autoimmune conditions frequently associated with PA, such as autoimmune thyroid disease, type I diabetes, and vitiligo, as part of the autoimmune polyendocrine syndromes (Eisenbarth & Gottlieb, 2004).

Albeit the *primum movens* of PA is AG, less frequently the disease is suspected due to symptoms of the gastrointestinal tract. The reason for the apparent paradox may lay in the fact that AG is associated with hypochlorhydria, and symptoms of the upper gastrointestinal tract are often related to the presence of chlorhydric acid. However, hypochlorhydria itself may cause impaired gastric emptying, eventually leading to dyspeptic symptoms such as epigastric discomfort, postprandial bloating and fullness, and early satiety (Annibale et al, 2005; Tosetti et al, 2000). Often the awareness and/or the concern about upper gastrointestinal symptoms and/or neurological symptoms are not sufficient to seek medical advice for this reason and specific data about clinical presentation in PA are scarce. In previous works performed on 177 consecutively diagnosed PA patients, dyspeptic symptoms were complained by 27.7%, and neurological symptoms, such as paresthesias, were present in 18.6% of patients, suggesting that the presence of these symptoms should be related to the possible

subclinical presence of PA (Annibale et al, 2000; Marignani et al, 1999). Anyway, in a very recent analysis PA patients were more frequently reporting no gastrointestinal symptoms, which were however present in half of them (47.5%) (Carabotti et al, 2017).

In a previous work, autoimmune thyroid disease showed a prevalence of nearly 40% in atrophic body gastritis patients (Lahner et al, 2008). The diagnosis of a concomitant presence of autoimmune thyroiditis and PA may have an important clinical implication, in particular in those patients who require replacement therapy with thyroxine, because patients with an impaired acid secretion may present thyroxine malabsorption requiring an increased dose of the drug (Lahner et al, 2014), and in patients with PA an associated hypochlorhydria is always present due to the loss of oxyntic mucosa (Annibale et al, 2011).

Useful information about in whom PA may be suspected, may derive also from epidemiological data. According to the older literature, PA is thought to be particularly common among individuals of Scandinavian, English, and Irish ancestry, while it appears to be much less common in Caucasians of Italian or Greek origin (Friedlander, 1934); in recent years, the disease have been reported in black and Latin-American subjects (Carmel, 1996; Carmel et al, 1978). In the so-called high-risk groups, about 9 new cases are detected per 100,000 population per year, and about 0.13% of the population is affected (Pedersen et al, 1969). The only more recent population survey reported that 1.9% of persons more than 60 years old has undiagnosed PA (Carmel, 1996).

A female preponderance ranging from 1.7 to 2.0:1 has been reported in white subjects (Babior, 1998). This gender distribution was confirmed in the more recent population survey of persons more than 60 years old conducted in California, in which the prevalence of PA was 2.7% in women and 1.4% in men (Carmel, 1996).

Regarding the age, PA is frequently described as a disease of adults >60 years of age (Carmel, 1996). In spite of this, some data challenged the common notion that PA is a disease of the elderly (Hershko et al, 2006), suggesting that in

clinical practice PA is probably underdiagnosed not only in elderly but also in younger patients.

Considering the clinical scenario, it may be reminded that (i) iron deficiency is a consequence of achlorhydria and may precede the development of PA (1) and (ii) iron deficiency may be present in concomitance with PA (Carmel et al, 1986). It has been shown that stratification by age cohorts from younger than 20 years to older than 60 years of patients with AG identified by hypergastrinemia and positive parietal cell antibodies showed a regular and progressive increase in mean corpuscular volume and levels of ferritin and gastrin and a decrease in vitamin B_{12} levels, whereas the prevalence of *H pylori* infection decreased from more than 80% at age younger than 20 years to 12.5% at age older than 60 years (Hershko et al, 2006). These findings support the idea that PA seems to be a disease starting many years before the establishment of clinical vitamin B_{12} deficiency.

Pernicious anemia: diagnosis and management

PA is defined as the presence of a hemoglobin concentration <13 g/dL for men and <12 g/dL for women (WHO, 2001), mean corpuscular volume (MCV) $\geq$100 fL (9), low levels of cobalamin (vitamin B_{12}) (Babior, 1998), together with the concomitant presence of AG and intrinsic factor deficiency.

Thus, by definition, PA is associated with AG, and strict diagnostic criteria for AG are based on the histological evidence of gastric body mucosal atrophy and ECL hyperplasia associated with hypochlorhydria to pentagastrin stimulation (Lee et al, 2002). Increased levels of fasting gastrin and decreased levels of Pepsinogen I are well accepted serological markers suggesting the presence of oxyntic mucosa damage (Kekki et al, 1991; Vaananen et al, 2003), which should be confirmed, however, by appropriate histological sampling of gastric body mucosa to definitively diagnose AG.

As far as regards gastric mucosa histology, corpus-restricted atrophy with a spared antrum is a classical and indispensable feature required for the diagnosis of PA. As reported in a previous review (Lahner & Annibale, 2001), in about 50% of PA patients antral mucosa is not spared, and in about 27% of PA

patients a concomitant antral atrophic gastritis may be observed. These data strongly suggest that an extension of gastritis to the gastric antrum does not necessarily exclude the diagnosis of PA and the presence of gastric autoimmunity. Also the determination of ECL cells hyperplasia is considered helpful in the histological diagnosis of AG associated with PA, because the presence of this histological change indirectly proves the presence of hypochlorhydria, which leads to hypergastrinemia which, in turn, is a trophic factor for ECL cells leading to their hyperplasia and eventually to the development of gastric carcinoids (Lahner et al, 2009).

Intrinsic factor deficiency would be proven by the now obsolete Schilling test: in order to confirm that the cobalamin deficiency is the result of intestinal malabsorption due to intrinsic factor deficiency, urinary excretion of orally administered vitamin B_{12} is low, and is increased by administration of vitamin B_{12} together with intrinsic factor. Unfortunately, the availability of this test is vanishing due to problems related to its radioactive reagents. In alternative, a gastric juice intrinsic factor assay has been proposed, which assesses the gastric intrinsic factor output after pentagastrin stimulation considering a value lower than 200 U/ h as diagnostic for PA (Cattan, 2011). However, probably because needing gastric tubage, this invasive test it has not earned wide popularity.

Therefore, in clinical practice, the presence of intrinsic factor deficiency may not be definitely proven and increasing reliance is placed on the detection of intrinsic factor antibodies for the diagnosis of PA, which are viewed as useful markers of this disease (Toh et al, 2007). Earlier studies reported the positivity to intrinsic factor antibodies in 40-60% of patients with PA (Ungar et al, 1967; Davidson et al, 1989), rising to 60-80% with increasing duration of disease (Carmel, 1992). In more recent years, the diagnostic performance of intrinsic factor and parietal cell antibodies has been assessed in patients with atrophic body gastritis with respect to cobalamin deficiency by using a novel ELISA assay (Lahner et al, 2009): in PA patients, intrinsic factor antibodies achieved a sensitivity and a specificity of 37% and 100%, respectively, and parietal cell antibodies yielded a sensitivity and a specificity of 81.5% and 90.3%, respectively. The combined assessment of both autoantibodies significantly

increased their diagnostic performance, yielding a 73% sensitivity for PA, while maintaining a 100% specificity. Thus, by combining the assessment of intrinsic factor and parietal cell autoantibodies the diagnostic performance of these surrogate markers for PA may notably be improved. Further, beyond as a specific hallmark of PA, the positivity to intrinsic factor and parietal cell antibodies may be interpreted also as an expression of oxyntic mucosa damage, because the increasing histological score of body mucosa atrophy correlated positively with the titer of both antibodies (Annibale et al, 2001; Annibale et al, 2005). The anemia seems to develop longitudinally over many years in anti-parietal cell antibodies-positive patients, symptomless, slowly promoting atrophy of the gastric mucosa and parietal cells (Rusak et al, 2016). In a very recent work conducted on atrophic gastritis patients, positivity to both ATP4A and ATP4Bautoantibodies resulted to be closely associated with atrophic body gastritis, suggesting that these assays should be valuable screening tools for detecting biomarkers of damaged atrophic oxyntic mucosa (Lahner et al, 2017).

Notwithstanding, an accurate differential diagnosis of other causes of cobalamin deficiency is mandatory. As previously reviewed (Lahner & Annibale, 2009), cobalamin deficiency may be caused by other causes of impaired absorption in the stomach or in the intestine, such as gastrectomy, ileal disease or resection, or by decreased intake due to vegetarianism. Among maldigestion, there are very rare cases related to severe pancreatic insufficiency, but more interesting is the recent evidence of maldigestion of dietary cobalamin in patients with corpus predominant *H pylori* gastritis leading to impaired acid secretion and consequent increased intragastric pH (Carmel et al, 1994; Cohen et al, 2000). In fact, dietary cobalamin is bound to salivary proteins, which need to be cleaved in presence of chlorhydric acid before it can be bound to intrinsic factor and be absorbed in the terminal ileum (Lahner & Annibale, 2009). In these cases of dietary cobalamin maldigestion Schilling test would be normal, indicating that cobalamin deficiency is not due to intrinsic factor deficiency. Without performing a Schilling test, it may be challenging to discriminate between the presence of PA and the presence of maldigestion of dietary cobalamin. However, from a practical point of view, the

clinical management of these two groups of patients is similar. Further, an accurate differential diagnosis should be carried out also for macrocytic anemia, which may underly other causes such as folate deficiency and myelodysplasia (Lahner & Annibale, 2009), and the assessment of serum homocysteine levels is be helpful. In this context, it should be kept in mind that in order to diagnose vitamin B_{12} deficency, total vitamin B_{12} measurement is used cost effectively as the parameter of choice, but it has limited sensitivity and specificity, especially in persons with vitamin B_{12} concentrations in the lower reference range (<400 pmol/L). In alternative, modern biomarkers for early diagnosis of vitamin B_{12} deficency, as holotranscobalamin (holoTC), also known as active B_{12}, and methyl malonic acid as a functional B_{12} marker have been proposed (Herrmann & Obeid, 2008). **Figure 1** shows the diagnostic clues for PA (Annibale et al, 2011).

The clinical management of patients with PA concerns two different aspects. First, the treatment of cobalamin deficiency and the monitoring of onset of iron deficiency. Second, the surveillance to early detect long-term consequences of PA, such as gastric cancer, carcinoids and, eventually, other type of cancers, with this last topic being the object of the present work.

The cobalamin replacement treatment is able to correct the anemia, whereas the neurological complications may be corrected only if the replacement treatment is given soon after their onset. However, the practice of giving cobalamin as an intramuscular injection has several drawbacks, because injections can be painful, difficult to provide for elderly or alone-living patients, and expensive if provided by health professionals. Thus, at least since the early 90ies, the usefulness of oral cobalamin treatment in PA has been debated (Hathcock et al, 1991; Lederle, 1991). According to a systematic review conducted by French hematologists (Lederle, 1991), several prospective studies (n= 4), prospective randomized studies (n= 3) and a systematic review by the Cochrane group (n=1) provide evidence that oral cobalamin therapy may adequately treat cobalamin deficiency, particularly hematological abnormalities or manifestations. These studies suggest that at least 1000 µg/day of oral cyanocobalmin are needed for pernicious anemia and a mean daily dose of 250

μg for food-cobalamin malabsorption, confirming the previously reported efficacy of oral cobalamin treatment in adult and elderly patients.

Pernicious anemia between autoimmunity and *H. pylori* infection

Atrophic body gastritis associated with PA is often called autoimmune gastritis or type A gastritis, which is defined as a type of chronic atrophic gastritis restricted to the body mucosa, characterized by a severe, diffuse atrophy of the oxyntic glands and hypochlorhydria, and a normal antral mucosa. Another classical histological feature of AG is the absence of *H pylori* on gastric mucosal biopsies (Lee et al, 2002). It is now accepted, that long-standing *H pylori* infection is able to induce atrophy of the gastric mucosa, and *H pylori* is considered the main causative agent of multifocal atrophic gastritis, in which the antrum is almost invariably involved (Peterson et al, 2002). Thus, AG is generally considered a separate entity from *H pylori*–related atrophic gastritis, mainly because the prevalence of *H pylori* infection in patients with severe AG and PA has been described to be low (Varis et al, 1993; Annibale et al, 2000). However, in the last years, the question has been raised whether *H pylori* may be implicated in the pathogenesis of AG, and, as basic mechanism for the induction of gastric autoimmunity by *H pylori* infection, molecular mimicry has been proposed (D'Elios et al, 2004; Appelmelk et al, 1998; Field et al, 2005). Molecular mimicry is defined as the possibility that sequence similarities between foreign and self-peptides are sufficient to result in the cross-activation of autoreactive T or B cells by pathogen-derived peptides and is a phenomenon associated with some pathogens where the antigens evoking an immune response have enough similarity to the body's own proteins to cause an autoimmune reaction, such as in rheumatoid arthritis, mediated by cross-reactive T cells and/or circulating antibodies. In fact, gastric H^+/K^+-ATPase has been recognized as the major autoantigen in experimental and human AG (Toh et al, 1992; Ma et al, 1994; Claeys et al, 1998), and autoreactive gastric CD4+T cells recognizing H+, K+-ATPase and *H pylori* antigens have been recently described in AG (Amedei et al, 2003; D'Elios et al, 2005). Thus, PA and AG seem to be an example of pathogen (*H pylori*)-induced-organ-specific autoimmunity,

in which genetical susceptibility plays an important role in relation to the loss of immunological tolerance (D'Elios et al, 2005). In fact, the immunological basis of molecular mimicry lies in the recognition by the T-cell antigen receptor of an antigenic peptide bound to a human leukocyte antigen (HLA) molecule on the surface on an antigen-presenting cell, and inappropriate activation of T cells may occur as a result of the up-regulation of HLA molecules in genetically susceptible individuals (Albert et al, 1999). A specific HLA-DR pattern has been suggested in PA patients several years ago (Ungar et al, 1981), and, more recently, blocking experiments with anti-DR and anti-DQ antibodies could show that DR probably represents the HLA restriction element in AG (Amedei et al, 2003).

The presence of *H pylori* infection was diagnosed by histology in up to 30% (median 11%), but by serology (IgG) in up to 51% (median 20.5%) of PA patients. It is well known that the diagnosis of *H pylori* infection may be difficult in patients with AG. *H pylori* may disappear over time due to the hostile gastric microenvironment, and past infection may be evidenced by positivity to *H pylori* serology in a large majority of patients with AG or PA (Annibale et al, 2000; Valle et al, 1999; Annibale et al, 2001). Another paper reported that the seropositivity against *H pylori* antigens may be evidenced in a very high percentage of patients with AG by using an *ad hoc* immunoblotting (Annibale et al, 2007): in this study 47.8% of AG patients had PA and all but two of them presented seropositivity against *H pylori* antigens including CagA and VacA. As far as regards histological features of the gastric body, in the vast majority of PA patients (>70%) this disorder is associated with severe body atrophy and the presence of intestinal metaplasia. Irrespective from the presence of *H pylori* infection, in about half of PA patients the gastric antrum is involved, and in about one third even by an antral atrophic gastritis, whose presence is strongly related to *H pylori* infection (Peterson et al, 2002). This observation challenges the widely accepted notion that PA occurs exclusively in association with the classical histological feature of corpus-restricted atrophic gastritis. All these data taken together supports the idea that long-standing *H pylori* infection probably plays an important role in many genetically susceptible PA patients,

in whom the active infectious process has been gradually replaced by an autoimmune process run by autoreactive gastric CD4+T cells recognizing H+, K+-ATPase and *H pylori* antigens, finally terminating in a burned-out infection and the irreversible destruction of the gastric body mucosa. The failure to evidence *H pylori* infection in some of these individuals does not necessarily argue against the role of the bacterium in these patients but, more likely indicates that a point of no return may be reached beyond which the autoimmune process may no longer require the continued presence of the inducing pathogen (Rose et al, 1993).

Pernicious anemia, autoimmune atrophic gastritis and risk of gastric neoplasms

Gastric cancer is still the fourth most common cancer worldwide and the second cause of cancer-related death (Lahner et al, 2015). A varying progression rate of atrophic gastritis to gastric cancer up to 2% per year has been reported at follow-up periods ranging from 1 to 16 years (Whiting et al, 2002; Dinis-Ribeiro et al, 2004; Vannella et al, 2010). A recent systematic review showed in atrophic gastritis patients with pernicious anemia a pooled gastric cancer incidence-rate of 0.3% person-year and an estimated 7-fold relative risk of gastric cancer (Vannella et al, 2013).

In patients with atrophic gastritis also type 1 gastric carcinoids may arise. Data on long-term incidence of type 1 gastric carcinoids are scanty (Annibale et al, 2001; Kokkola et al, 1998; Sjoblom et al, 1988). A recent cohort study reported an annual incidence rate for type 1 gastric carcinoid of 0.4% (Vannella et al, 2011), while an older study reported an annual incidence of 2%, observing 8 new cases of type 1 gastric carcinoids in 416 patient-year (Kokkola et al, 1998). In the above cited study, pernicious anemia was present in almost 50% of patients with type 1 gastric carcinoids (Vannella et al, 2011), while previous studies included exclusively patients with this condition (Kokkola et al, 1998; Sjoblom et al, 1988; Sjoblom et al, 1993; Armbrecht et al, 1990; Stockbrugger et al, 1983).In patients with atrophic gastritis, the need and cost-effectiveness of regular endoscopic follow-up for gastric cancer surveillance is not definitely

established. Recent European guidelines recommend a scheduled surveillance for gastric cancer for those patients who have extensive – i.e. both antrum and gastric body –atrophic gastritis or intestinal metaplasia (Dinis-Ribeiro et al, 2012). However these guidelines are not addressed to patients with pernicious anemia, as corpus-restricted atrophic gastritis with antrum-spared, typically present in pernicious anemia patients, is not considered to be part of the precancerous cascade described by Correa (Correa et al, 1992). According to the data reported, different clinical management of atrophic gastritis patients with or without pernicious anemia does not seem to be justified, raising questions whether these recommendations should include also pernicious anemia patients.

With regard to surveillance for type 1 gastric carcinoids, indications are even more uncertain. A recent study on endoscopic management of these tumours, reported that for atrophic gastritis patients without recurring type 1 gastric carcinoids, endoscopic controls might be planned yearly in the early follow-up, but can probably become less intensive with endoscopic controls every 4 years according to atrophic gastritis screening for gastric cancer risk (Merola et al, 2011). To better evaluate the value of surveillance in atrophic gastritis patients and establish follow-up frequencies, more precise data on the occurrence of gastric neoplastic lesions, preferably obtained in large prospective studies with adequate follow-up, are needed (de Vries et al, 2007).

The combined risk of gastric cancer and carcinoids together has been investigated many years ago limited to pernicious anemia patients (Kokkola et al, 1998; Sjoblom et al, 1988). A recent study (Lahner et al, 2015) investigated in a prospective cohort of patients with atrophic gastritis the occurrence of gastric cancer and carcinoids at long-term follow-up from 4 years upwards. In this study a total of 200 atrophic gastritis patients from a prospective cohort (67% females, median age 55 years) with a follow-up of 7.5 (range 4-23.4) years were included. Follow-up gastroscopies at 4-years intervals were performed. The results of this study showed that, overall, 22 gastric neoplastic lesions were detected (crude incidence 11%). Gastric cancer was diagnosed in 4 patients at a median follow-up of 7.2 years (crude incidence 2%). Eleven type I-gastric

carcinoids were detected at a median follow-up of 5.1 years (crude incidence of 5.5%). In 7 patients, 6 low-grade and 1 high-grade dysplasia were found. The annual incidence rates person-year were 0.25%, 0.43%, and 0.68% for gastric cancer, dysplasia, and type 1 gastric carcinoids, respectively. From this study emerged that in atrophic gastritis patients at long-term follow-up an annual incidence rate of 1.36% person-year for gastric neoplastic lesions and that the incidence rates of gastric cancer and type 1 gastric carcinoid were not different (p=0.07), indicating that atrophic gastritis patients are similarly exposed to both risks (Lahner et al, 2015). According to Globocan 2012, the annual incidence rate for gastric cancer in the general Italian population is estimated to be 0.004% (GLOBOCAN, 2012). This study thus provides further evidence confirming the increased risk for gastric cancer in atrophic gastritis. The presence of pernicious anemia is associated with gastric carcinoids but not with gastric cancer, as survival free of carcinoids is significantly shorter in patients with pernicious anemia.

The patients' features associated with gastric cancer and type 1 gastric carcinoids are different, keeping in step with the different pathogenetic mechanisms of these two type of tumors (Delle Fave et al, 2012). The occurrence of type 1 gastric carcinoids is mainly associated with features of autoimmune gastritis as pernicious anemia and positivity to gastric autoantibodies. Gastric cancer, instead, is associated with the presence of *H. pylori* in the corporal mucosa (HR 8) (Lahner et al, 2015) keeping in step with the concept of corpus-predominant gastritis as observed by Uemura more than ten years ago that *H. pylori* positive patients and those with severe gastric atrophy, corpus-predominant gastritis or intestinal metaplasia are at increased risk for gastric cancer (Uemura et al, 2001). The presence of pernicious anemia is associated with gastric carcinoids but not with gastric cancer, as survival free of carcinoids is significantly shorter in patients with pernicious anemia.

Atrophic patients are exposed to a double risk of gastric neoplastic lesions, gastric cancer and type 1 gastric carcinoids: In a retrospective case-series (Lahner et al, 2015) the occurrence of gastric cancer in patients with type 1 gastric carcinoids was described in 23% (4 out of 17) of patients with type 1

gastric carcinoids over a median follow-up period of 6 years. Three cases were intestinal-type adenocarcinomas and one a signet-ring cells diffuse gastric cancer, localized in 3 cases in the antrum. Thus, it seems to be worthwhile to monitor type 1 gastric carcinoids patients by a long-term surveillance programme, including an accurate bioptic sampling of antral mucosa. The effects of long-standing hypergastrinemia may be a possible explanation why patients with type 1 gastric carcinoids might develop more frequently gastric cancer. Hypergastrinemia has been proposed in many models of gastric carcinogenesis and seems to be a common causative factor in otherwise different circumstances; in all species where long-term hypergastrinemia has been induced, an increased risk of gastric malignancy, with adenocarcinoma phenotype and even the signet-ring cells phenotype, was observed (Waldum et al, 2014; Fossmark et al, 2008). Moreover, the long-term conservative management of type 1 gastric carcinoids exposes these patients to a higher risk of gastric cancer. This risk is basically present in atrophic gastritis, due to the pathophysiological changes related to gastric body atrophy, such as increased pH, reduced ascorbic acid and scavenging of nitrites and other potential carcinogenic substances (Fossmark et al, 2008).

It has become apparent that besides *H. pylori,* other bacteria may be involved in gastric carcinogenesis; it has been shown that the gastric cancer microbiota was dominated by species of the genera *Streptococcus, Lactobacillus, Veiollonella* and *Prevotella*, albeit the roles of these species in the development of gastric cancer needs to be determined (Walker et al, 2014).

Although the gastric cancer incidence has declined over the past decades, especially in Western countries, the mortality rate due to gastric cancer remains high (Ferlay et al, 2013). Detection and surveillance of patients with premalignant conditions, as atrophic gastritis and intestinal metaplasia, could potentially lead to detection and treatment of advanced lesions – i.e dysplastic lesions and gastric cancer – in an early stage (de Vries et al, 2008; Gonzalez et al, 2010; den Hoed et al, 2013). In patients with premalignant conditions, the risk of developing gastric cancer may be further stratified by the location, severity, and

extent of gastric atrophy and/or metaplasia (El-Zimaity, 2006; Leung et al, 2004).

Several histological classifications have been developed for atrophic gastritis and preneoplastic changes. To date, in clinical practice and in research, the updated Sydney System is mainly used. This system combines topographic, morphological, and etiological information to standardize histological reporting (Dixon et al, 1996). More recently, the systems known as OLGA (operative link for gastritis assessment), and OLGIM (operative link on gastric intestinal metaplasia) assessment have been proposed for staging of gastritis (Rugge & Genta, 2005). Unfortunately, classifications are still difficult to use in clinical practice, and often present the disadvantage of important inter- and intra-observer variation. (Rugge et al, 2011).

As mandatory conditions to correctly adopt premalignant gastric lesions as reliable indicators for gastric cancer development, a standardized biopsy sampling protocol and an uniform, reproducible histological grading system need to be applied in clinical practice (Dinis-Ribeiro et al, 2012). A recent nationwide survey investigated in a community-based endoscopic setting what really happens in clinical practice with regard to the detection of gastric atrophy and intestinal metaplasia in dyspeptic patients (Lahner et al, 2014). In detail, a nationwide survey was conducted on 979 consecutive patients (50–65 years old) with dyspeptic symptoms, who were examined at24 gastrointestinal endoscopy units throughout Italy. Clinical information was collected from questionnaires; a standard bioptic mapping was performed in each unit, biopsies from each patient were analyzed by histopathology performed according to daily clinical practice in each local pathology centre. The results showed that separate descriptions of antral and corporal biopsies were included in 679 pathology reports (69%), whereas the standardized Sydney system was applied in 324 reports (33%). Gastric atrophy without intestinal metaplasia and gastric atrophy with intestinal metaplasia were detected in 322 (33%) patients. The full adherence to Sydney system significantly increased the probability of detecting gastric atrophy with intestinal metaplasia (OR 9.6, 95% CI 5.5-16.7), gastric atrophy without intestinal metaplasia (OR 1.92, 95% CI 1.07-3.44), and either of

the conditions (OR 6.67, 95% CI 4.36-10.19). Thus, according to these findings, in daily routine practice only one third of histology reports were worked out adhering to Sydney system showing that international guidelines are poorly observed in clinical practice (Lahner et al, 2014). This may represent a critical element for surveillance strategies for gastric cancer.

Pernicious anemia and risk of extra-gastric and non-gastrointestinal neoplasms

In addition to gastric cancer and type 1 gastric carcinoid, pernicious anemia may also be associated with an increased risk of other cancers (**Table 1**). The role for immune dysregulation in hematological malignancies is well established; however, little is known about immune-related diseases, such as pernicious anemia, and the risk of hematological malignancies. Several works investigated for a possible association between pernicious anemia and hematological malignancies. Overall, pernicious anemia seemed to increase the risk for multiple myeloma (Brown et al, 2008; Anderson et al, 2009a; Murphy et al, 2015), acute myeloid leukemia and myelodisplastic syndrome (Anderson, 2009b; Murphy et al, 2015). Conversely, the association between pernicious anemia and multiple myeloma was not confirmed by Lewis et al and Lindqvist et al. The study from Soderberg et al did not find any association between pernicious anemia and all hematological diseases, as well.

Hypergastrinemia has been identified in patients with pernicious anemia as a physiologic response to the damage to the oxyntic mucosa and achlorhydria. However, beyond the role of gastrin in gastric acid secretion, it was also shown to stimulate the growth of epithelial cells and prevent apoptosis (Boursi et al, 2016). Gastrin and its precursors activate several pathways important in tumorigenesis such as the beta catenin, MAP kinase and JAK2/STAT3 pathways and were described in association with colorectal, pancreatic and liver cancers, other than gastric cancers and carcinoids (Talley et al, 1989; Borch et al, 1988; Karlson et al, 2000). Recently, Murphy et al found that individuals with pernicious anemia seem to be at increased risk for tonsilar cancer, hypopharyngeal cancer, esophageal squamous cell carcinoma, small intestinal

cancer and liver cancer. Both studies from Murphy et al and Boursi et al found no association between pernicious anemia and colorectal cancer risk.

Previous studies have lacked the large number of cancer cases necessary to precisely estimate the magnitude of the association of pernicious anemia with other type of cancers. This paucity of prior studies has resulted in conflicting evidences and in the lack of any guidelines of clinical management for patients with pernicious anemia.

AIM OF THE STUDY

Therefore, the aim of the present study was to systematically review the available literature on PA and the development of gastrointestinal cancers other than gastric cancers (GI-other than GC) and non-gastrointestinal cancers (non-GIC) to estimate their incidence-rates.

METHODS AND MATERIALS

Literature search strategy

The search was conducted according to PRISMA guidelines (Moher et al, 2009). The electronic database PubMed MEDLINE (U.S. National Library of Medicine, Bethesda, MD) was systematically searched combining as main keywords "pernicious anemia", "autoimmune gastritis", "cancer".

The search was translated into the following query: ("pernicious anaemia"[All Fields] OR "anemia, pernicious"[MeSH Terms] OR ("anemia"[All Fields] AND "pernicious"[All Fields]) OR "pernicious anemia"[All Fields] OR ("pernicious"[All Fields] AND "anemia"[All Fields])) OR (autoimmune[All Fields] AND ("gastritis"[MeSH Terms] OR "gastritis"[All Fields])) AND ("neoplasms"[MeSH Terms] OR "neoplasms"[All Fields] OR "cancer"[All Fields]) AND ("humans"[MeSH Terms] AND (English[lang] OR French[lang] OR German[lang] OR Spanish[lang])).

No publication date restrictions were imposed. Only reports published in English, German, French, Italian, and Spanish language were considered.

Study selection

Studies published up to April 27th 2017 were included in the systematic review if they fulfilled all of the following criteria: 1) observational study including patients with PA and reporting the numbers of non-gastric and non-GI cancers identified during a defined follow-up period; 2) the study was performed in adult patients; 3) the study was an original full paper which presented unique

data. Studies were excluded if 1) they were reviews, letters, editorials, case-reports; 2) follow-up data were not available.

Potentially relevant articles were screened for eligibility independently in an unblinded standardized manner by the two reviewers (MC, EL), initially by abstract and then by full text when necessary to determine whether they met the inclusion criteria. Disagreement between reviewers was resolved by discussion. The reference lists of the identified articles as well as of identified relevant reviews were manually searched for additional studies that may have been missed using the computer-assisted search strategy.

Data Extraction

Two reviewers (MC, EL) independently extracted the following information from each publication: the first author's last name, year of publication, country of study location, study design, criteria for diagnosis of PA, sources of selection of participants, numbers of patients investigated, duration of follow-up period (years), calculation of person-years, age of patients (median or mean or range), gender, methods for identification of cancer, type of cancers, numbers of cancer cases.

Outcome

The outcome measure of interest was the incidence-rate of GI-other than GCs and non-GICs, which were calculated as the ratio between numbers of new cancer cases identified during the follow-up period and PA patients.

Statistical analysis

We extracted from each included study the number of incident non-GC and non-GIC cases and the number of PA patients exposed to risk. The cumulative incidence and the person-years (PY) incidence-rates in PA patients were calculated by the reviewers if not explicitly stated.

The cumulative incidence was calculated as ratio between new non-GC and non-GIC cases and the number of PA patients. The PY incidence rates were calculated as the ratio between new non-GC and non-GIC cases and person-

years. The pooled PY incidence rates of non-GC and non-GIC cases per organ or systems were calculated as the ratio between sum of new cases and the sum of PY. These incidence rates were then used and compared by meta-analysis with the annual cancer incidence rates of both genders in the general population aged over 40 years as reported for the single European countries by the website GLOBOCAN (GLOBOCAN, 2012). The weighted summary proportion (pooled proportion) under the fixed and random effects model were calculated by Freeman-Tukey transformation. In case of a positive "Heterogeneity test" (p-value <0.05) the more appropriate random effects model was taken into consideration in which both the random variation within the studies and the variation between the different studies is incorporated (DerSimonian et al, 1986). The extent of heterogeneity was investigated by Cochran's Q and I^2 statistic. Q is the weighted sum of squares on a standardized scale, with low p-values indicating presence of heterogeneity. I^2 is the percentage of observed total variation across studies that is due to real heterogeneity rather than chance, calculated as $I^2 = 100\% \times (Q - df)/Q$ (where Q is Cochran's heterogeneity statistic and df the degrees of freedom. I^2 lies between 0% and 100%, a value of 0% indicates no observed heterogeneity, larger values show increasing heterogeneity (Higgins et al, 2003).

The statistical analysis was carried out using a dedicated software package (MedCalc Software, Mariakerke, Belgium, version 12.7.8.0).

Quality Assessment

The two reviewers evaluated the quality of all included studies using the Newcastle-Otawa quality assessment scale (Wells et al). This scale awards a maximum of five points to each study. The considered categories are randomization, blinding of outcome assessment, description of withdrawals and dropouts, description and appropriateness of randomization and blinding. A study can be awarded a maximum of one point for each category. Discrepancy in quality assessment was discussed and resolved by two reviewers.

RESULTS

Search results

The electronic search strategy identified a total of 1.150records from electronic database (**Figure 2**). These articles were screened on the basis of title and abstract and, after application of the inclusion and exclusion criteria, 97 articles were retrieved for full-paper evaluation. Of these 97 full-papers, 20 met eligibility criteria and were subjected to data extraction (Armbrecht et al, 1990; Arvanitakis et al, 1979; Blackburn et al, 1967; Borch et al, 1986, Brinton et al, 1986; Chan et al, 2008; Demmlet et al, 1966; Elsborg et al, 1973; Fallah et al, 2014; Hemminki et al, 20112; Hoffman et al, 1970; Hsing et al 1993; Landgren et al, 2011; Mellemkjaer et al, 1996; Mosbech et al, 1950; Shah et al, 20124; Siurala et al, 1966; Talley et al, 1989; von Knorre et al, 1975; Ye et al, 2003). Manual searching of reference lists of potentially relevant papers and reviews added 4 articles.

Quality Assessment

Details of the quality assessment of included studies are given in **Table 2**. Of the twenty included studies, four (20.0%) (Borch, Demmler, Siurala, von Knorre) resulted of very low quality, five (25.0%) (Armbrecht, Arvanitakis, Chan, Elsborg, Hoffman) of low quality, six (30.0%) (Blackburn, Fallah, Hemminki, Mosbech, Talley, Ye) of medium qualities and five (25.0%) (Brinton, Hsing, Landgren, Mellemkjaer, Saha) of high quality according to the Newcastle-Ottawa quality assessment scale.

Characteristics of Included Studies

The characteristics of the included studies are summarized in **Table 3**. Female gender was prevalent in 9 studies with a range between 51.3% and 72.7%, 44.3% in 1 study, only male patients were investigated in 2 studies, while in the remaining studies gender was not reported. Patients had a median age of 71.4 years, in all studies ranging from 56.4 (Mosbech) to 79.0 (Chan) years.

The majority of the 20 studies were European, in particular 9 (45.0%) were from Northern Europe (Denmark, Finland, Sweden) and 5 (25.0%) from other European countries (United Kingdom, Germany). Five studies (25.0%) were American and only one study (5.0%) was performed in Eastern Countries (China). The design was prospective in 10 (50%) studies. Diagnostic criteria for the diagnosis of PA were carefully described in 10 (50.0%) studies. In particular, clinical criteria assessed by blood tests (macrocytic anemia, low serum vitamin B_{12}or iron levels, hypergastrinemia, low pepsinogen I levels), positive Schilling test, gastric pH or megaloblastic bone marrow were used in 9 studies (Armbrecht, Arvanitakis, Borch, Chan, Hoffman, Mosbech, Siurala, Talley, von Knorre); data were extracted on the basis of International Classification of Disease (ICD) in 7 studies (Brinton, Hemminki, Hsing, Landgren, Mellemkjaer, Shah, Ye) and using hospital records in one study (Fallah). Three studies (Blackburn, Demmler, Elsborg) included patients with PA without providing further details. Most of the studies included hospitalized patients for PA, that could be after followed-up as outpatients, and 3 studies included outpatients referred to hematological units for anemia or local healthcare providers (Shah, Siurala, Talley). In the Blackburn study, patients were recorded in the files of eight PA clinics, but it was not indicated whether the patients were hospitalized or not.

Most of the studies retrieved the diagnosis of GI and non-GI cancers biopsies using ICD codes from National Registers, three from local computer-based files (Chan, Landgren, Talley), while 4 studies made use of clinical interview/death certificates (Blackburn, Mosbech, Siurala, von Knorre). Three studies did not specified how cancer diagnoses were assessed (Demmler, Elsborg, Hoffman).

Regarding cancer incidence, 4 studies reported only GI-other than GC cases (Shah, Talley, von Knorre, Ye) and 3 studies only non-GIC cases (Elsborg, Fallah, Siurala), while the remaining studies reported both of them.

Cancer incidence

With regard to the cancer incidence in PA patients included in this analysis, we found that the cumulative incidence-rate was 2.4%, ranging from 0.2%(Ye) to 15.8% (Arvanitakis), with 1,993 new cancer cases on a total PA population of 82,257 patients (15,285,818.3 PY) (**Table 3**), corresponding to a cumulative incidence rate of 13.0(100,000-PY). In particular, cancer incidence rates ranged from 0.2% (Shah, Ye) to 9.2% (Hsing) in the 9 Northern European studies, from 0.2% (Shah) to 8.3% (von Knorre) in the 5 studies of other European countries, from 5.1% to 15.8% in the 5 American studies and was 6.0% in the Chinese study.

Overall, a total of 67,654 (9,392,404.8 PY) and 45,373 (6,825,006.7 PY) PA patients were studied, with 767 and 1,194new cancer cases in GI-other than GC and non-GIC group, respectively.

Looking at GI-other than gastric cancers, the incidence rate was 1.1% (8.2 100,000-PY), ranging from 0.2% (Shah, Ye) to 8.0% (Borch) (**Table 4**). Looking at non-GI cancers, the incidence rate was 2.6% (17.5 100,000-PY), ranging from 0.3% (Hemminki) to 13.2% (Arvanitakis) (**Table 5**).

Heterogeneity between studies was statistically significant in both groups ($p<0.0001$), with Chi-square test=761.02 and degree of freedom=14 in the GI-other than GC group and Chi-square test=1730,21 and degree of freedom=15 in the non-GIC group. A meta-analysis was performed as shown in **Figure 3A** and **3B** by considering random and fixed effects, respectively, given the significance level of heterogeneity. We found a pooled incidence rate per person-year for non-GCs and non-GICs of 0.27 (95%CI: 0.16-0.42] and 0.23 [95%CI: 0.22-0-25].

Analysing the cancer incidence per organ or system (**Table 6**) (**Figure 4**), we found that the highest incidence rates (100,000-PY) were observed for skin cancer (32.8 – driven by non melanoma cancer: 61.6) and prostatic cancer (31.9). A meta-analysis performed using the calculated annual cancer incidence rates per organ showed an overall relative risk for cancer in PA as 0.68 (95% CI: 0.48-0.95). With regard to site-specific cancers, PA patients (RR: 95%CI) showed a lower RR as compared to the general population for lung (0.26: 0,21 to 0,31), thyroid (0.34: 0,21 to 0,57), esophagus (0.26: 0,178 to 0,371), colo-rectum (0.14:

0,098 to 0,19), liver (0.18: 0,13 to 0,24), pancreas (0.65: 0,45 to 0,93), breast (0.17: 0,13 to 0,22), ovary (0.45: 0,30 to 0,67), prostate (0.72:0,623 to 0,84), kidney (0.61: 0,43 to 0,86) and non-melanoma skin cancer (0.56: 0.50-0.61). In contrast, PA patients showed a higher risk than the general population for biliary tract cancer (1.81: 1,21 to 2,70) and for hematological malignancies as multiple myeloma (2.83: 1,76 to 4,55), Hodgkin lymphoma (3.0: 1,35 to 6,68), non Hodgkin lymphoma (2.08: 1,58 to 2,75), and leukemia (1.56: 1,16 to 2,12) (**Figure 5**).

DISCUSSION

Evidence on overall cancer incidence in PA are quite sparse and conflicting. To our knowledge, this is the first systematic review on the estimate of GI-other than GCs and non-GICs incidence in these patients.

From 1,154 articles retrieved from a search strategy with no time limit, we selected 20 papers according with our inclusion criteria, published between 1950 and 2014 concerning a total of 82,257 PA patients (15,285,818.3 PY).

Overall, 1,993 new cases of cancer other than gastric were identified, corresponding to an overall incidence rate of 2.4%, ranging from 0.2% to 15.8%. In detail, 767 and 1,194 new cases of GI cancers other than GC and non-GIC, respectively, were identified, with a crude incidence rate of 1.1% (8.2100,000-PY) and 2.6% (15.7100,000-PY), respectively. Comparing with the general population, PA patients showed an overall lower relative risk (0.68) of developing cancer, even if we found an increased risk for haematological malignancies (in particular, multiple myeloma [2.83] and Hodgkin lymphoma [3.0]), and biliary tract cancers (1.8).

Individuals with pernicious anemia have long been suggested to have increased risk of gastric cancer as well as gastric carcinoid tumors in a number of studies. Such an association is plausible, as a less acidic stomach (hypochlorhydria) as well as chronic inflammation are among the mechanisms by which *H. pylori* is thought to cause gastric cancer. In addition to gastric cancer, pernicious anemia has been reported to be associated with an increased risk of other cancers.

Anyways, the paucity of prior studies of PA and cancer leave this association still controversial.

In our analysis, we found that PA patients showed a lower relative risk of GI-other than GCs (esophagus, liver, pancreas and colorectum). The possible association between PA and GI-other than GCs has been quite debated in literature, though providing conflicting evidence. The inverse relationship between gastric atrophy and esophageal cancer has already been observed in a small number of studies (Cook et al, 2010; Islami et al, 2011; Kamangar et al, 2009), which is thought to reflect reduced acid reflux from the stomach to the esophagus as a result of the loss of parietal cell. On the contrary, Murphy et al found a positive association with esophageal squamous cell carcinoma, but no association with esophageal adenocarcinoma. As the authors stated, this relationship is not fully understood and may, or may not, be causal (Murphy et al, 2015). Gastrin at physiological concentrations was shown to act as a growth factor for colorectal cancer both through endocrine as well autocrine/paracrine mechanisms and pre-malignant adenomas were also shown to express an isoform of the cholecystokinin B/gastrin receptor (Singh et al, 2000; Smith et al, 2000). However, the lack of association between PA and colorectal cancer risk was recently demonstrated in the largest population study currently available (Boursi et al, 2016). Only a previous work showed an increased risk of liver cancer for people with PA (Hsing et al, 1993), even if it was recently confirmed only in women (Murphy et al, 2015). No specific association of PA with lung, breast, prostate and kidney cancer has been reported in a recent population-based study (Murphy et al, 2015), supporting our results. Thus, the finding of the present systematic review keep in step with these data denying the higher risk of developing cancers in these organs in patients with PA.

In our analysis, we found an increased relative risk for haematological malignancies and biliary tract cancers. The role for immune dysregulation in hematological malignancies is well established and several works investigated for a possible association between PA and hematological malignancies. Overall, PA seemed to increase the risk for multiple myeloma (Anderson et al, 2009a; Brown et al, 2008; Murphy et al, 2015), acute myeloid leukemia and

myelodisplastic syndrome (Murphy et al, 2015, Anderson et al2009b). Conversely, the association between pernicious anemia and multiple myeloma was not confirmed by Lewis et al and Lindqvist et al. The study from Soderberg et al did not find any association between PA and all hematological diseases, as well.

The significant association between the risk of haematological malignancies could be related to vitamin B12 deficiency (Anderson et al, 2009a; Anderson et al, 2009b). Vitamin B12 deficiency could promote the development of cancer by causing derangement of one-carbon metabolism nutrients (folate, vitamin B6, riboflavin, homocysteine) associated to aberration in both DNA methylation and DNA synthesis, as proposed by several authors (Friso & Choi, 2005; Miranti et al, 2017). When insufficient B12 is available, cellular folates accumulate in the methylfolate form, but cannot be metabolically utilized due to the block in the methyonine synthase reaction, creating a functional folate deficiency commonly referred to as "methylfolate trap" (Herbert, 1994). Previous studies in bone marrow cells and immortalized cultured cells (Wickramasinghe & Fida, 1994; Wickramasinghe & Fida, 1993) demonstrated that B12 deficiency increased uracil misincorporation into DNA, suggesting that this functional folate deficiency by B_{12} deficiency results in inadequate thymidylate synthesis. Some authors observed that localized B_{12} deficiency in squamous-cell lung cancer tissue was associated with genomic DNA hypomethylation, suggesting that decreased availability of methylfolate by B_{12} deficiency subsequently reduces genomic DNA methylation (Friso & Choi, 2005).

We also found a positive association of PA with the risk of biliary tract cancers. There are no specific literature data on this association, anyways, a possible role for DNA hypomethylation can be hypothesized, considering that aberrant DNA methylation is an epigenetic mechanisms that can occur early in carcinogenesis (Esteller et al, 2008). A very recent molecular study demonstrated the up-regulation of Dicer in cholangiocarcinoma cells, promoting their proliferation and invasion (Cheng et al, 2017). Dicer is known to regulate methylation of CpG island in mammalian cancer cells (Ting et al, 2008, Chen et al, 2011), interacting

with heterocromatin protein 1α (HP1α). DNA methylation has also become one of the key molecular mechanisms in the tumorigenesis of gallbladder (Tekcham et al, 2016). In a very recent study, the aberrant methylation of the WIF-1 promoter was found to be involved in the malignant transformation of gallbladder cancer (Lin et al, 2017). Also, hypergastrinemia has been identified in PA patients as a physiologic response to the damage of the oxyntic mucosa and achlorhydria. However, beyond the role of gastrin in gastric acid secretion, it was also shown to stimulate the growth of epithelial cells and prevent apoptosis (Todisco et al, 2001; Seva et al, 1994; Ferrand et al, 2006; Maddalo et al, 2014).

We are aware that this study has some limits. To evaluate the quality of included studies we applied the Newcastle-Ottawa Quality Scale for cohort studies, which permits a critical appraisal of data (Well et al). We found that almost half of the included studies had high/medium quality. In spite of this, clinical and methodological heterogeneity was present among the 20 studies included, resulting in a wide range for the observed overall cancer incidence rate (0.2%-15.8%). The different approach across the studies should take into account the decade in which the studies were performed. Diagnostic tools for both PA and cancer diagnosis have changed over time. A cancer diagnosis obtained from death certificates in the older studies could be not directly comparable with those obtained from ICD code in National Cancer Registers. Also, 6 six studies did not specifically report how cancer was detected. Moreover, we excluded from this analysis the incidence rate for gastric cancer. We know that the introduction of the fiberscope and, subsequently, of the video endoscope has been a revolution in the diagnosis of stomach diseases. This observation could impact the number of esophageal cancers reported in older studies: some of those cases could include primary misdiagnosed gastric cancer. Notwithstanding these limits, we performed a meta-analysis based on the calculated GI-other than gastric and non-GI cancer incidence rates in PA patients. A further limit of this systematic review is that for the comparison with the general population used for the calculations of the relative risk of cancer in PA data from cancer registries were used, but the retrieved study did

not report on controls and this was a way to get estimates on the association of the single cancers with PA. Though we are aware that the results of this meta-analysis may be biased by the limits explained above, we think however that they should be acceptable as an estimate of the overall probability of PA patients to develop cancer and may represent a starting point to design further population-based prospective studies. Of note, this is the first systematic review performed on this topic.

In conclusion, this systematic review showed an overall cancer risk for PA patients that do not appear to be increased in comparison to the general population, except for haematological malignancies and biliary tract cancers, except for haematological malignancies and biliary tract cancers. Further high quality studies are needed to confirm the risk of specific cancer development in PA patients, so as to provide clinical guidance for a careful follow-up of these patients.

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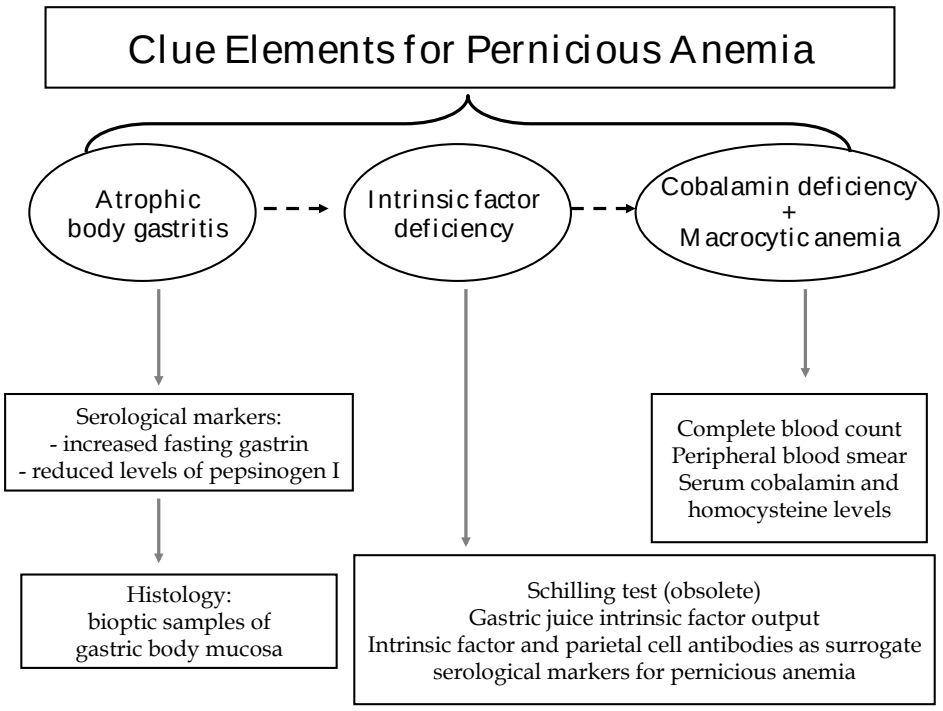


Figure 1 – Diagnosis of pernicious anemia (Annibale et al, 2011).

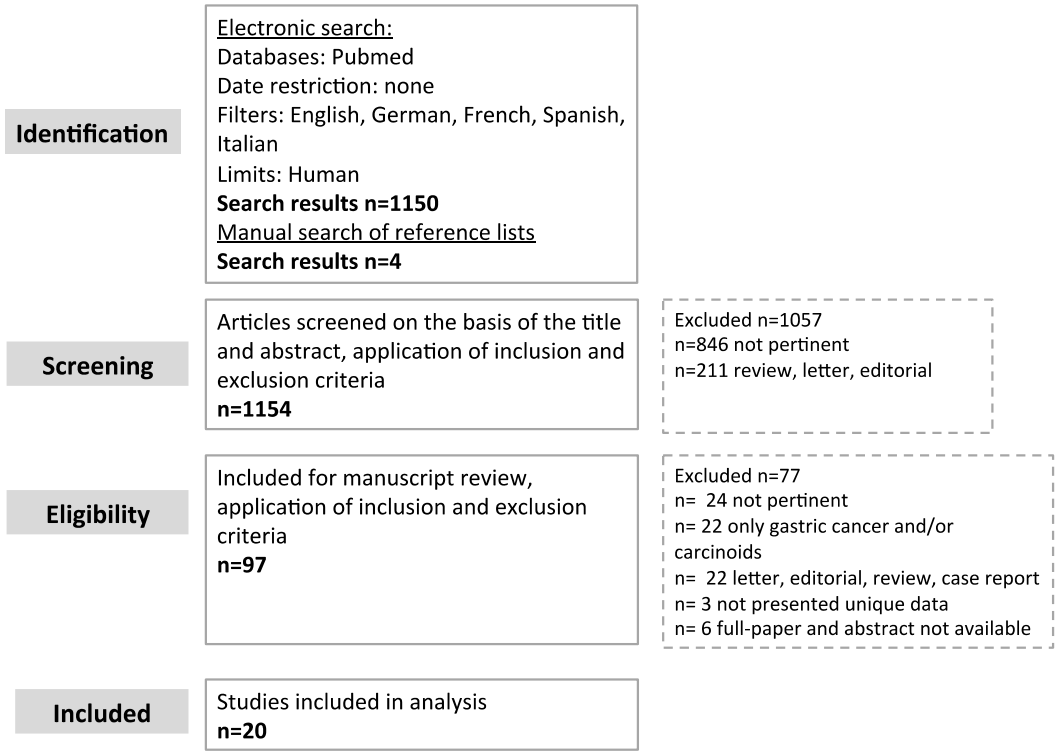
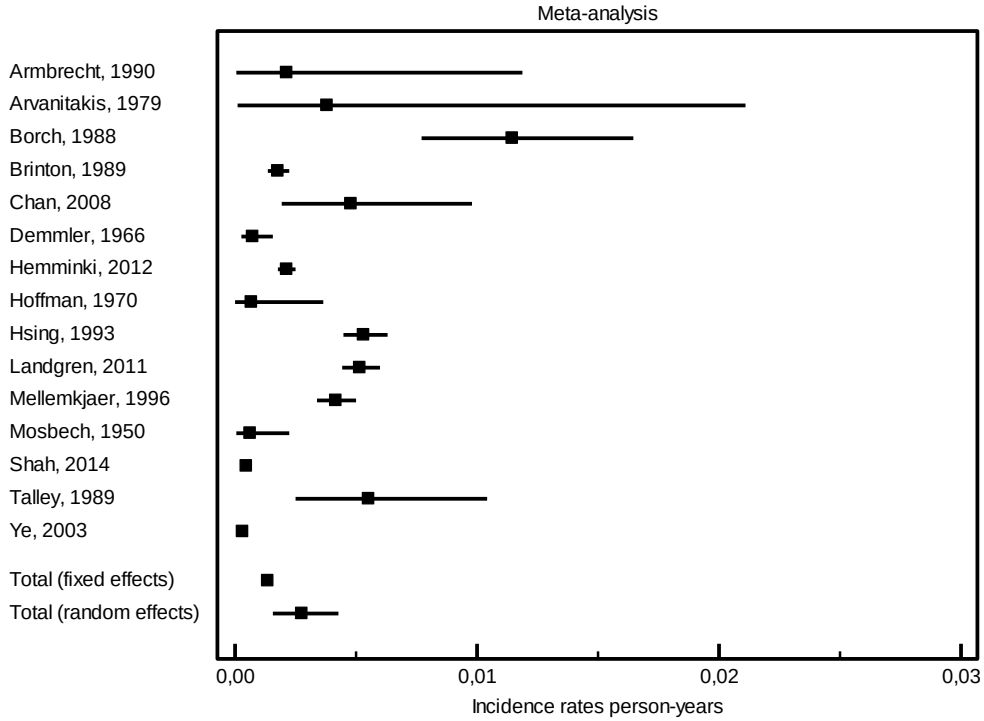


Figure 2 – Flow-chart of study selection.

A



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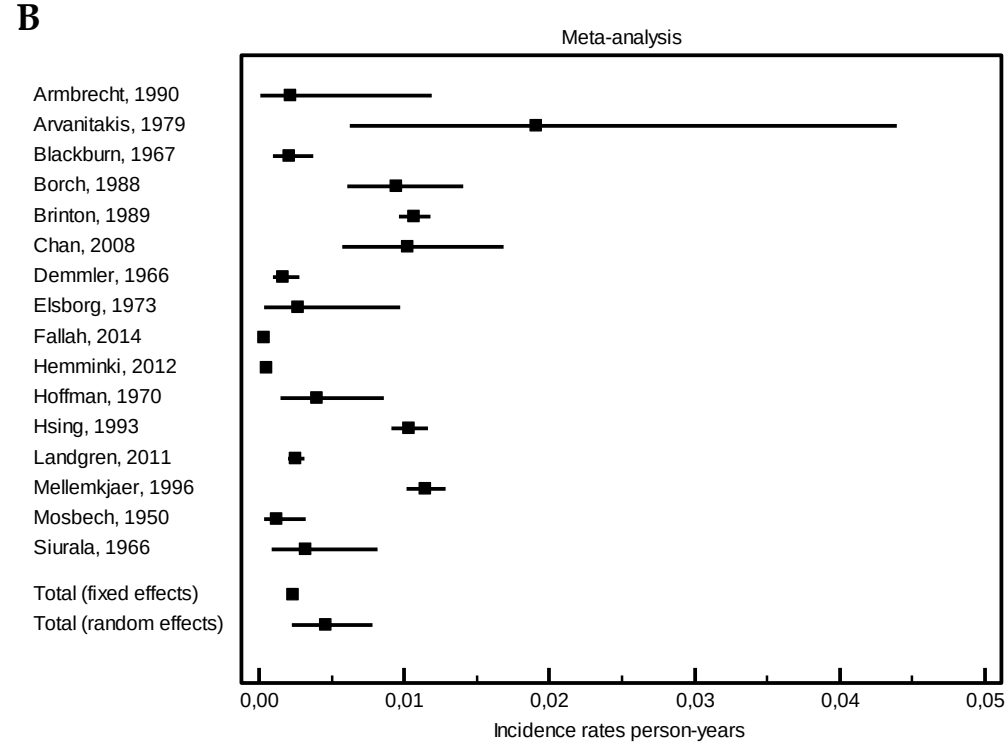

Figure 3 – Annual cancer incidence rates in PA patients of the selected studies. A) GI-other than gastric cancer incidence. p<0.0001, I^2=98.16% (95%CI 97.68-98.54)**; B) Non-GI cancer incidence.** p<0.0001, I^2=99.13% (95%CI 98.97-99.27)**.**

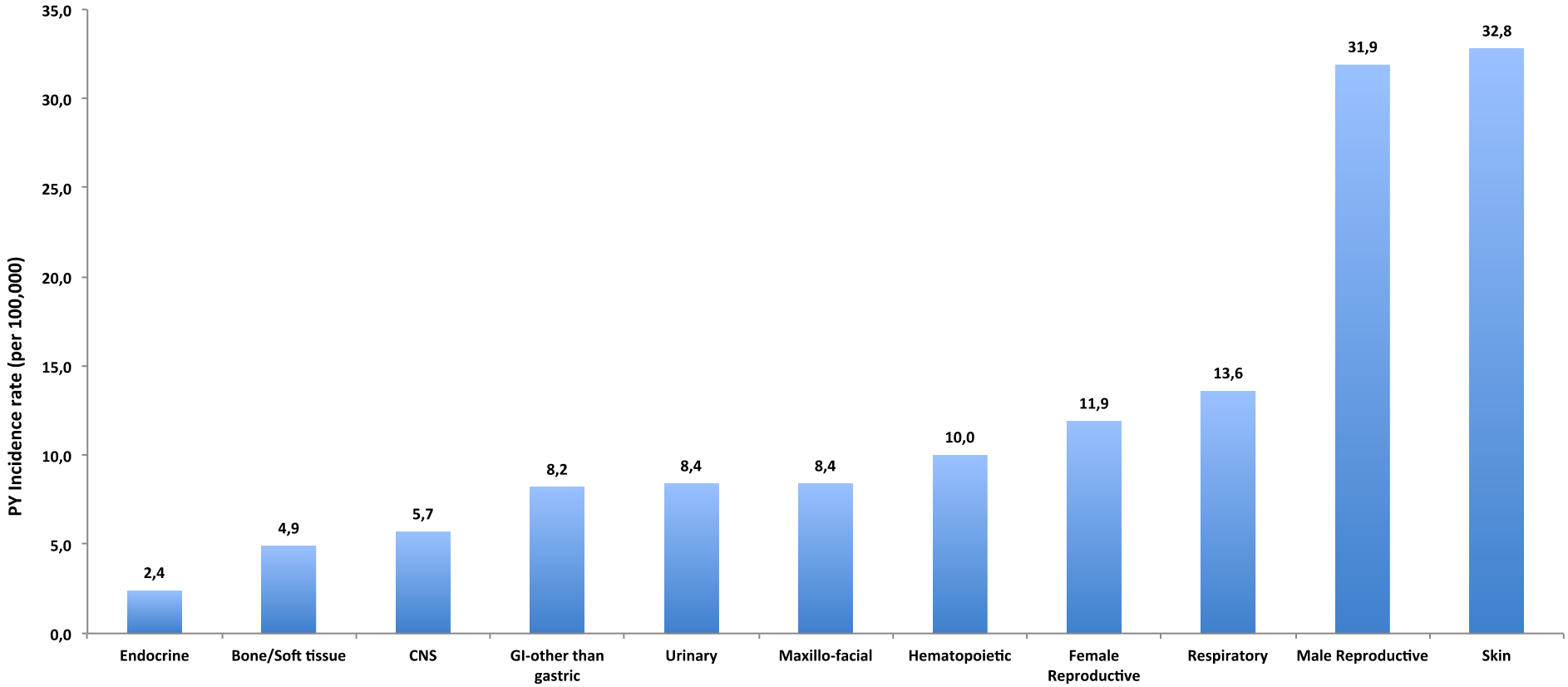

Figure 4 - Cancer incidence (PY per 100,000) per organ/system in PA patients.

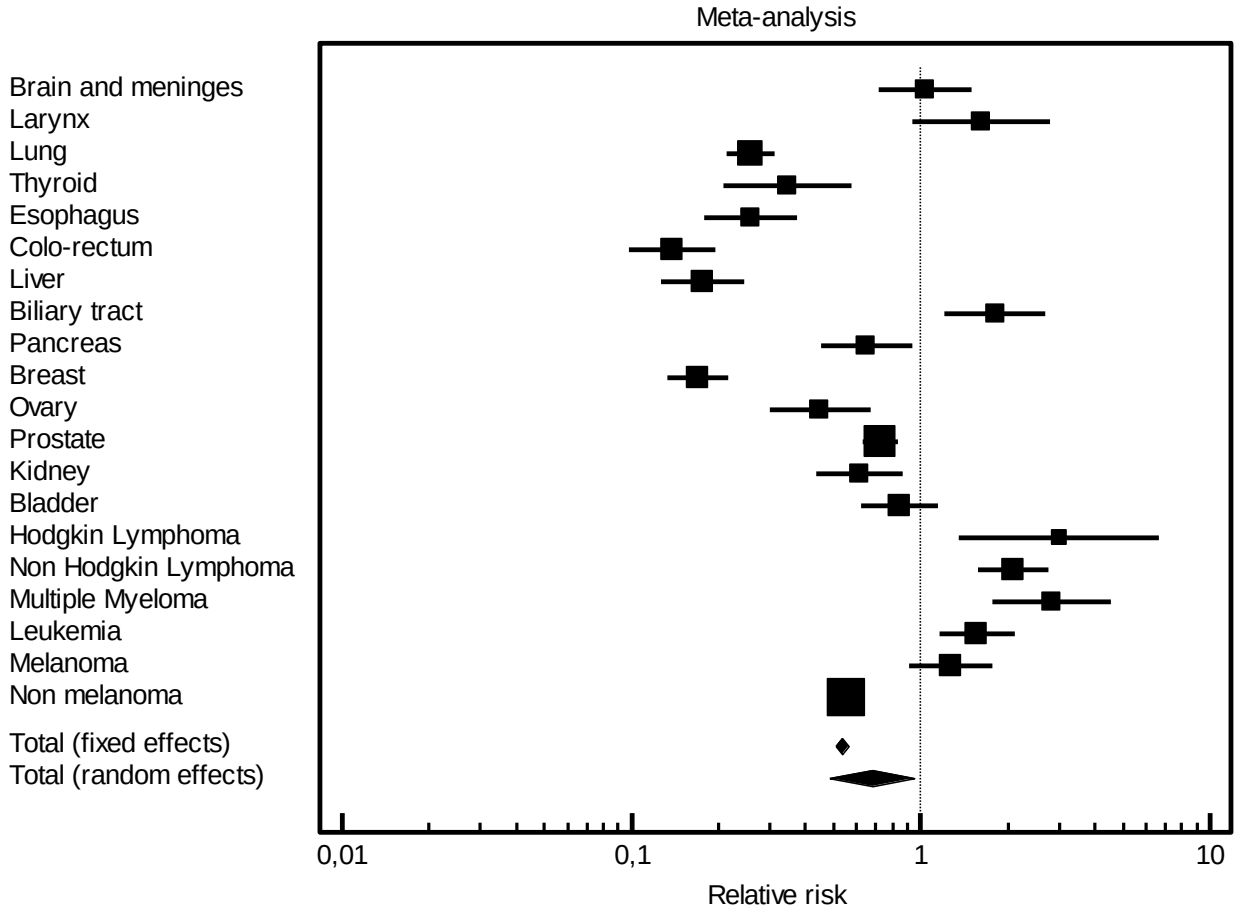

Figure 5 – Cancer relative risk per organ in PA patients comparing with the general population. p<0.0001; I^2=96.79%; 95% CI: 95.92-97.47.

Author	Country	Study Design	PA Diagnosis	Patients (inpatients, outpatients, general population)	Cancer Cases (n)	Age, yrs Median or Mean or Range (Female, %)	Cancer type	Cases of cancer with pernicious anemia, n (OR)
Anderson, 2009	US	Population-based case-control study	Hospital, physician and outpatient Medicare claims	Patients from SMAHRT (hematopietic Malignancy Risk Traits) study using SEER-Medicare data	44.350	67-84 (51.4%)	Lymphoid malignancies	Non Hodgkin Lymphoma=565 (1.0) Hodgkin Lymphoma=11 (0.6) Multiple Myeloma=222 (1.5)
Anderson, 2009	US	Population-based case-control study	Hospital, physician and outpatient Medicare claims	Patients from SMAHRT (hematopietic Malignancy Risk Traits) study using SEER-Medicare data	13.486	77 (50.9%)	Myeloid malignancies	Acute myeloid leukemia=177 (1.57) Chronic myeloid leukemia=21 (0.74) Myelodysplastic syndromes=148 (2.38) Chronic myeloproliferative disorder=25 (1.11)
Boursi, 2016	UK	Nested case-control study	Read code	Patients traced from THIN databse	22.098	71.3 (45%)	Colorectal cancer	154 (1.02)
Brown, 2008	US	Retrospective cohort study	ICD code	Patients traced from discharge records in US Veterans Affairs Hospitals	4.641	59.9 (0%)	Multiple myeloma (MM)/MGUS	MM= 27 (RR 2.15) MGUS= 14 (RR 1.97)
Landgren, 2006	Sweden, Denmark	Retrospective population-based study	ICD code from Swedish Inpatient Register and Danish Inpatient and Outpatient Register	Patients traced from the Swedish Family-Cancer Database and the Danish Cancer Register and Danish Central Population Register	7.764	54-76 (30-35%)	Chronic lymphocitic leukemia (CLL)	CLL=31 (1.94)
Lewis, 1994	US	Population-based, case-control study	Personal interviews	Patients traced from 3 US Population-based cancer registries	573	62.3-65.3 (47.4-55.8%)	Multiple myeloma (MM)	MM=11 (1.1)
Lindqvist, 2011	Sweden	Retrospective, population case-control study	Discharge diagnosis from Swedish Inpatient Register	Patients raced from Swedish Cancer Register and Swedish Inpatient Register	7.314	71 (45-50%)	Multiple myeloma (MM)/MGUS	MM=48 (1.2) MGUS=19 (1.8)

Murphy, 2015	US	Population-based, case-control study	Medicare claims data	Patients traced from SEER database-Medicare	1.138.390	65->85 (47%)	All type of cancer	Tonsil (2.00) Hypopharinx (1.92) Small bowel (1.63) Liver (1.49) Myeloma (1.55) Acute myeloid leukemia (1.68) Myelodysplastic syndrome (2.87)
Soderberg, 2006	Sweden	Case-control study	Swedish Hospital Discharge Registry	Patients traced from the Swedish Hospital Discharge Registry and the Swedish Cancer Registry	ns	ns	Hematological malignancies	Chronic Lymphoid Leukemia (CLL)=8 (0.7) Acute Myeloid Luekemia=7 (1.1) CML=1 (0.6) Leukemia (exlc. CLL)=14 (1.3) Non Hodking Lymphoma=29 (0.8) Myeloma=14 (0.8) All hematological malignancies=66 (0.9)

Table 1 - Pernicious anemia in cancer patients. PA: pernicious anemia; ns: not specified.

Author Year	Selection				Comparability		Outcome assessment			Global Quality Assessment
	1	2	3	4	1	2	1	2	3	
Armbrecht 1990	*		*	*			*	*	*	LQ
Arvanitakis 1979		*	*		*		*	*		LQ
Blackburn 1967	*	*	*		*	*	*		*	MQ
Borch 1986	*		*	*			*	*	*	VLQ
Brinton 1989	*	*	*	*	*	*	*	*	*	HQ
Chan 2008	*		*				*	*	*	LQ
Demmler 1966	*		*				*	*		VLQ
Elsborg 1973	*		*				*	*	*	LQ
Fallah 2014	*	*	*		*		*	*	*	MQ
Hemminki 2012	*	*	*		*		*	*	*	MQ
Hoffman 1970	*		*	*			*	*	*	LQ
Hsing 1993	*	*	*	*	*	*	*	*	*	HQ
Landgren 2011	*	*	*	*	*	*	*	*	*	HQ
Mellemkjaer 1996	*	*	*	*	*	*	*	*	*	HQ
Mosbech 1950	*	*	*		*	*	*	*	*	MQ
Shah 2014	*	*	*	*	*	*	*	*	*	HQ
Siurala 1966		*					*		*	VLQ
Talley 1989		*	*	*	*	*	*	*		MQ
Von Knorre 1975	*		*				*	*		VLQ
Ye 2003	*	*	*	*	*	*	*	*		MQ

Table 2 – Quality Assessment of included studiesaccording to the Newcastle-Otawa quality assessment scale. HQ= high quality (nine stars); MQ=medium quality (seven or eight stars); LQ=low quality (five or six stars); VLQ=very low quality (four stars or less).

Author	Country	Study Design	PA Diagnosis	Patients (inpatients, outpatients, general population)	Population (n)	Age, yrs Median or Mean or Range (Female, %)	Follow-up, years (person-years)	Diagnostic Tool for Identification of cancer	Cases of GI-other than GC, n (incidence, %)	Cases of non-GIC, n (incidence, %)	Overall cases of cancers other than gastric, n (incidence, %)
Armbrecht, 1990	Germany	Prospective, follow-up study	BT BM	Patients traced from departments of Medicine, Haematology, Gastroenterology in London	73	63 (20-79) 44.3 %	6,4 (467,2)	ns (symptoms monitoring during clinical follow-up)	1 (1.4%)	1 (1.4%)	2 (2.7)
Arvanitakis, 1979	USA	Prospective	BT ST BM Ach	ns	38	69.4 (44-88) 51.3%	6.9 (1-43) (262.2)	ns (symptoms monitoring during clinical follow-up)	1 (2.6%)	5 (13.1%)	6 (15.8)
Blackburn, 1967	United Kingdom	Prospective	nr	Patients recorded in the files of eight PA clinics	1625	nr	3 (4,875)	Death certificates	44 (2.7%)	10 (0.6%)	54 (3.3)
Borch, 1988	Sweden	Prospective	BT ST	Patients from two healthcare districts	361	73 (66.2%)	7 (2,527)	National Cancer Register and Regional Oncologic Center records - histologically verified	14 (3.9%)	15 (4.1%)	29 (8.0)
Brinton, 1989	USA	Retrospective cohort-analysis	ICD on computer-based file	Patients hospitalized at Veterans Administration Hospitals, across USA	5161	72.8 0%	6.8 (35,094.8)	ICD on computer-based file	62 (1.2%)	375 (7.3%)	437 (8.5)

Chan, 2008	China	Hospital-based, longitudinal prospective study	BT BM	Patients hospitalized at single hospital in Hong Kong	367	79 (57-93) nr	≈4 (≈1,468)	Electronic patient-record system	7 (1.9%)	15 (4.0%)	22 (6.0)
Demmler, 1966	Germany	Retrospective	nr	Patients hospitalized in 35 German Hospitals	238	nr	maximum 35 years (≈8,330)	ns	6 (2.5%)	14 (5.9%)	20 (8.4)
Elsborg, 1973	Denmark	Prospective	nr	Patients hospitalized for PA and followed-up as outpatients	82	68 (33-86) 61%	9 (0.5-31)* (738)	ns	0	2 (2.4%)	2 (2.4)
Fallah, 2014	Sweden	Retrospective cohort study	Hospital discharges, Outpatients registries, Primary Health Care registries	Patients traced from Swedish healthcare databases	12.651	nr	9.4 (118,919.4)	Records from Swedish Cancer Registry and cause-specific death registry	-	45 (0.3%)	45 (0.3)
Hemminki, 2012	Sweden	Follow-up study	ICD code at hospitalization	Patients traced from the Swedish Hospital Discharge Register (hospitalized PA) and Swedish Cancer Registry	11.839	ns	5.8 (68,666.2)	Records from Swedish Cancer Registry	145 (1.2%)	34 (0.3%)	179 (1.5)
Hoffman, 1970	USA	Prospective	BT ST	Patients hospitalized for PA and followed-up as outpatients	138	74 (33-87) nr	≈11 (≈1,518)	ns	1 (0.7%)	6 (4.3%)	7 (5.1)
Hsing, 1993	Sweden	Prospective	ICD on computer-based file	Patients hospitalized for PA in the Uppsala health	4517	73.1 55.3%	5.9 (26,650.2)	Records from the Swedish Cancer Registry in linkage with	142 (3.1%)	275 (6.1%)	417 (9.2)

				care region that includes 6 countries in Sweden				the Swedish National Death Register			
Landgren, 2011	USA	Retrospective population-based study	ICD on computer-based file	Patients hospitalized at Veterans Administration Hospitals, across USA	2810	57.67 0%	≈12.02 (≈33,776.2)	ICD code from discharge diagnosis	175 (6.2%)	85 (3.0%)	260 (9.2)
Mellemkjaer, 1996	Denmark	Retrospective	ICD on computer-based file	Patients traced from nationwide Hospital Discharge Register in Denmark	5072	71 for male; 73 for female; 66%	5.1 (1-15) (25,767.2)	ICD code from Danish Cancer Registry	107 (2.1%)	295 (5.8%)	402 (7.9)
Mosbech, 1950	Denmark	Retrospective	BT Ach	Patients hospitalized for PA and followed-up as outpatients	301	56.4 for male; 59.1 for female; 72.7%	10.3 for male, 10.7 for female (10.6) (3,190.6)	Death certificates	2 (0.7%)	4 (1.3%)	6 (2.0)
Shah, 2014	UK	Retrospective	THIN Read diagnostic code	Patients traced in The Health Improvement Network (THIN) database	15.324	68.79 - 69%	4.31 (66,046.44)	THIN Read diagnostic code	30 (0.2%)	-	30 (0.2)
Siurala, 1966	Finland	Prospective	BT Ach ST	Outpatients referred to Hematological Department	100	ns	10-15 (1,250)	Follow-up interview on death causes	0	4 (4%)	4 (4)
Talley, 1989	USA	Retrospective population-based cohort study	BT BM ST Ach	Patients traced from the Rochester Epidemiology Project database	150	70 (63%)	10.9 (1,635)	Outpatient and Inpatient records of local Medicare providers	9 (6%)	-	9 (6)
Von Knorre, 1975	Germany	Retrospective	BT BM Ach	Patients hospitalized for PA and followed-up as outpatients	145	nr	13.1 (2-35) (1,899.5)	Death Certificates	12 (8.3%)	-	12 (8.3)
Ye, 2003	Sweden	Retrospective register-based cohort study	ICD code in Swedish Inpatient Register	Patients traced from Swedish Inpatient Register linked to the Swedish Cancer Register	21.265	74.3 (60.3%)	7.1 (150,981.5)	ICD code from the Swedish Cancer Register	50 (0.2%)	-	)

Table 3 - Charact

eristics of the included studies. PA: pernicious anemia; ns: not specified; nr: not reported; *duration from the time of PA diagnosis; ST: Schilling test; BT: Blood test; BM: bone marrow; Ach: achloryd ria; EGD: gastrosco py.

Author	Country	Study Design	PA Diagnosis	Patients (inpatients, outpatients, general population)	Population (n)	Age, yrs Median or Mean or Range (Female, %)	Follow-up, years (person-years)	Cases of GI-other than GC, n (cumulative incidence, %) [incidence rate PY, %]
Armbrecht, 1990	Germany	Prospective, follow-up study	BT BM	Patients traced from departments of	73	63 (20-79) 44.3	6,4 (467.2)	**All=1 (1.4) [0.2]** Colon=1

				Medicine, Haematology, Gastroenterology in London				
Arvanitakis, 1979	USA	Prospective	BT ST BM Ach	ns	38	69.4 (44-88) 51.3	6.9 (1-43) (262.2)	**All=1 (2.6) [0.4]** Rectum=1
Borch, 1988	Sweden	Prospective	BT ST	Patients from two healthcare districts	361	73 (66.2)	7 (2,527)	**All=29 (8.0) [1.1]** Colon=4 Rectum=1 Liver=1 Biliary tract=1 Pancreas=5 §Other digestive=17
Brinton, 1989	USA	Retrospective cohort-analysis	ICD on computer-based file	Patients hospitalized at Veterans Administration Hospitals, across USA	5161	72.8 0	6.8 (35,095)	**All=62 (1.2) [0.2]** Esophagus=11 Small bowel=1 Colon=34 Rectum=1 Liver=2 Biliary tract=4 Pancreas=7 §Other digestive=2
Chan, 2008	China	Hospital-based, longitudinal prospective study	BT BM Neu	Patients hospitalized at single hospital in Hong Kong	367	79 (57-93) nr	≈4 (≈1,468)	**All =7 (1.9) [0.5]** Small bowel=1 Colon=4 Liver=1 Biliary tract=1
Demmler, 1966	Germany	Retrospective	nr	Patients hospitalized in 35 German Hospitals	238	nr	maximum 35 years (≈8,330)	**All=6 (2.5) [0.07]** Esophagus=1 Small bowel=1 Rectum=1 Pancreas=3
Hemminki, 2012	Sweden	Follow-up study	ICD code at hospitalization	Patients traced from the Swedish Hospital Discharge Register (hospitalized PA) and Swedish Cancer Registry	11.839	ns	5.8 (68,666.2)	**All=145 (1.2) [0.2]** Esophagus=26 Small bowel=9 Colon=72 Rectum=35 Anus=3
Hoffman, 1970	USA	Prospective	BT ST	Patients hospitalized for PA and followed-up as outpatients	138	74 (33-87) nr	≈11 (≈1,518)	**All=1 (0.7) [0.07]** Colon=1
Hsing, 1993	Sweden	Prospective	ICD on	Patients hospitalized for	4517	73.1	5.9	**All=142 (3.1) [0.5]**

			computer-based file	PA in the Uppsala health care region that includes 6 countries in Sweden		55.3	(26,650.3)	Esophagus=15 Colon=36 Rectum=25 Liver=14 Biliary tract=18 Pancreas=34
Landgren, 2011	USA	Retrospective population-based study	ICD on computer-based file	Patients hospitalized at Veterans Administration Hospitals, across USA	2810	57.67 0	≈12.02 (≈33,776.2)	**All=175 (6.2) [0.5]** Esophagus=30 Small bowel=5 Colon=59 Rectum=38 Liver=18 Pancreas=25
Mellemkjaer, 1996	Denmark	Retrospective	ICD on computer-based file	Patients traced from nationwide Hospital Discharge Register in Denmark	5072	71 for male; 73 for female; 66	5.1 (1-15) (25,768.2)	**All=107 (2.1) [0.4]** Esophagus=8 Small bowel=2 Colon=50 Rectum=19 Liver=3 Biliary tract=6 Pancreas=19
Mosbech, 1950	Denmark	retrospective	BT Ach	Patients hospitalized for PA and followed-up as outpatients	301	56.4 for male; 59.1 for female; 72.7	10.3 for male 10.7 for female (10.69 (3190.6)	**All=2 (0.7) [0.06]** Colon=1 Liver=1
Shah, 2014	UK	Retrospective	THIN Read diagnostic code	Patients traced in The Health Improvement Network (THIN) database	15.324	68.79 - 69	4.31 (66,046.44)	**All=30 (0.2) [0.04]** Pancreas=30
Talley, 1989	US	Retrospective population-based cohort study	BT BM ST Ach	Patients traced from the Rochester Epidemiology Project database	150	70 (63)	10.9 (1,635)	**All=9 (6.0) [0.6]** Colon=9

Ye, 2003		Retrospective register-based cohort study	ICD code in Swedish Inpatient Register	Patients traced from Swedish Inpatient Register linked to the Swedish Cancer Register	21.265	74.3 (60.3)	7.1 (150,981.5)	**All=50 (0.2) [0.03]** Esophagus=50
OVERALL					67.654		138.83 (9,392,404.82)	**ALL=767 (1.1) [0.008]**

Table 4 - Characteristics of the included studies reporting GI-other than gastric cancer cases. PA: pernicious anemia; ns: not specified; na: not applicable; nr: not reported; *duration from the time of PA diagnosis; §Cancer type not specified; ST: Schilling test; BT: Blood test; BM: bone marrow; Ach: achlorhydria; EGD: gastroscopy. 34 and 12 cancer cases reported from Blackburn (1967) and van Knorre (1975), respectively, were excluded because it was not specified if GI or extra-GI cases.

Author	Country	Study Design	PA Diagnosis	Patients (inpatients, outpatients, general population)	Population (n)	Age, yrs Median or Mean or Range (Female, %)	Follow-up, years (person-years)	Cases of non-GIC, n (cumulative incidence, %) [incidence rate PY, %]
Armbrecht, 1990	Germany	Prospective, follow-up study	BT BM	Patients traced from departments of Medicine, Haematology, Gastroenterology in London	73	63 (20-79) 44.3	6,4 (467.2)	**All=1 (1.4) [0.2]** Thyroid=1
Arvanitakis, 1979	USA	Prospective	BT ST BM Ach	ns	38	69.4 (44-88) 51.3	6.9 (1-43) (262.2)	**All=5 (13.2) [1.9]** CNS (Meningioma)=1 Sinus maxillary=1 Larynx=1 Thyroid=1 Kidney and lymphoma and acoustic neuroma=1
Blackburn, 1967	United Kingdom	Prospective	nr	Patients recorded in the files of eight PA clinics	1625	nr	3 (4,875)	**All=10 (0.6) [0.2]** Leukemia=9 Lymphosarcoma=1
Borch, 1988	Sweden	Prospective	BT ST	Patients from two healthcare districts	361	73 (66.2)	7 (2,527)	**All=24§ (6.6) [0.9]**
Brinton, 1989	USA	Retrospective cohort-analysis	ICD on computer-based file	Patients hospitalized at Veterans Administration Hospitals, across USA	5161	72.8 0	6.8 (35,094.8)	**All=375 (7.3) [1.1]** CNS=6 Eye=1 Thyroid=1 Buccal cavity and pharynx=50 Lung= 80 Other respiratory=13 Bone=6 Connective/soft tissue=5 Melanoma=12 Breast=1 Prostate=90 Other genital=3 Bladder=24 Kidney and other urinary=10

								Hodgkin Lymphoma=1 Other lymphoid tissue=6 Multiple myeloma=9 Lymphatic leukemia=4 Myeloid leukemia=17 Other leukemia=10 Other=26**§**
Chan, 2008	China	Hospital-based, longitudinal prospective study	BT BM Neu	Patients hospitalized at single hospital in Hong Kong	367	79 (57-93) nr	≈4 (≈1,468)	**All=15 (4.1) [1.0]** CNS (meningioma)=1 Hypopharynx=1 Lung=7 Soft tissue (head and neck)=1 Thymoma=1 Breast=1 Cervix uteri=1 Corpus uteri=1 Myelodisplastic syndrome=1
Demmler, 1966	Germany	Retrospective	nr	Patients hospitalized in 35 German Hospitals	238	nr	maximum 35 years (≈8,330)	**All=14 (5.9) [0.2]** Sinus maxillary=1 Lower lip=1 Lung=5 Breast=1 Ovarian=1 Cervix uteri=1 Kidney=1 Bladder=1 Connective (sarcoma)=1 Metastasis (unknown primary)=1
Elsborg, 1973	Denmark	Prospective	nr	Patients hospitalized for PA and followed-up as outpatients	82 61	68 (33-86)	9 (0.5-31)* (738)	**All=2 (2.4) [0.3]** Lung=1 Acute leukemia=1
Fallah, 2014	Sweden	Retrospective cohort study	Hospital discharges, Outpatients registries, Primary Health Care registries	Patients traced from Swedish healthcare databases	12.651	nr	9.4 (118,919.4)	**All=45 (0.4) [0.04]** Non Hodgkin lymphoma=45
Hemminki, 2012	Sweden	Follow-up study	ICD code at hospitalization	Patients traced from the Swedish Hospital Discharge Register (hospitalized PA) and Swedish Cancer Registry	11.839	ns	5.8 (68,666.2)	**All=34 (0.3) [0.05]** Upper digestive tract (pharynx)=34
Hoffman, 1970	USA	Prospective	BT ST	Patients hospitalized for PA and followed-	138	74 (33-87) nr	≈11 (≈1,518)	**All=6 (4.3) [0.4]** Prostate=3

				up as outpatients				Multiple myeloma=1 Hematologicsl malignancies=2
Hsing, 1993	Sweden	Prospective	ICD on computer-based file	Patients hospitalized for PA in the Uppsala health care region that includes 6 countries in Sweden	4517	73.1 55.3	5.9 (26,650.2)	**All=275 (6.1) [1.0]** CNS=10 Buccal cavity=16 Larynx=1 Lung=25 Prostate=58 Breast=36 Cervix uteri=6 Corpus uteri=10 Ovary=9 Kidney=16 Bladder=12 Melanoma=5 Non melanoma skin cancer=8 Lymphoma=11 Endocrine=7 Non Hodgkin Lymphoma=11 Hodgkin disease=2 Multiple Myeloma=11 Chronic Lymphoid leukemia=6 Myeloid leukemia=8 Other leukemia=7
Landgren, 2011	USA	Retrospective population-based study	ICD on computer-based file	Patients hospitalized at Veterans Administration Hospitals, across USA	2810	57.67 0	≈12.02 (≈33,776.2)	**All=85 (3.0) [0.2]** Buccal cavity=85

Mellemkjaer, 1996	Denmark	Retrospective	ICD on computer-based file	Patients traced from nationwide Hospital Discharge Register in Denmark	5072	71 for male; 73 for female 66	5.1 (1-15) (25,767.2)	**All=295 (5.8) [1.1]** CNS=7 Buccal cavity and pharynx=16 Lung=45 Breast=39 Cervix=1 Uterus=10 Ovary=6 Prostate=39 Kidney=14 Bladder=23 Melanoma=4 Non melanoma skin cancer=57 Non Hodgkin Lymphoma=14 Multiple myeloma=7 Leukemia=13
Mosbech, 1950	Denmark	retrospective	BT Ach	Patients hospitalized for PA and followed-up as outpatients	301	56.4 for male; 59.1 for female 72.7	10.3 for male 10.7 for female (3,190.6)	**All=4 (1.3) [0.1]** Thyroid=1 Breast=1 Cervix uteri=1 Prostate=1
Siurala, 1966	Finland	Prospective	BT Ach ST	Outpatients referred to Hematological Department	100		10-15 (1,250)	**All=4 (4.0) [0.3]** Thyroid=1 Lung=2 Leukemia=1
OVERALL					45,373		150,2 (6,825,006.66)	**ALL=1.194 (2.6) [0.02]**

Table 5 - Characteristics of the included studies reporting non-GI cancers. PA: pernicious anemia; ns: not specified; na: not applicable; nr: not reported; *duration from the time of PA diagnosis; §Cancer type not specified; ST: Schilling test; BT: Blood test; BM: bone marrow; Ach: achlorhydria; EGD: gastroscopy. 34 and 12 cancer cases reported from Blackburn (1967) and van Knorre (1975), respectively, were excluded because it was not specified if GI or extra-GI cases.

Organ System	Cases of cancer/PA patients (cumulative incidence %) [annual incidence rate per 100.000 person-years]	Organ	Cases of cancer/PA patients (cumulative incidence %)[annual incidence rate per 100.000 person-years]	References
CNS	25/15.155 (0.2) [5.7]	Brain and meninges	25/15.155 (0.2) [5.7]	1 Arvanitakis, 6 Brinton, 1 Chan, 10 Hsing, 7 Mellemkjaer
Maxillo-facial	206/30.042 (0.7) [8.4]	Sinus maxillary	2/276 (0.7) [17.3]	1 Arvanitakis, 1 Demmler
		Eye	1/1561 (0.06) [2.8]	1 Brinton
		Buccal cavity and pharynx	203/30.004 (0.7) [9.1]	50 Brinton, 1 Chan, 1 Demmler, 34 Hemminki, 16 Hsing, 85 Landgren, 16 Mellemkjaer
Respiratory	180/15.575 (1.2) **[13.6]**	Larynx	2/4.555 (0.04) [3.4]	1 Arvanitakis, 1 Hsing,
		Lung	165/15.537 (1.1) [13.6]	80 Brinton, 7 Chan, 5 Demmler, 1 Elsborg, 25 Hsing, 45 Mellemkjaer, 2 Siurala
		Other	13/5.161 (0.2) [37.0]	13 Brinton
Endocrine	12/10.190 (0.1) [2.4]	Thyroid	5/5.673 (0.1) [2.0]	1 Ambrecht, 1 Arvanitakis, 1 Brinton, 1 Mosbech, 1 Siurala
		Other	7/4.517 (0.1) [26.3]	7 Hsing
GI other than gastric	767/67.654 (1.1) [8.2]	Esophagus	141/50.902 (0.3) [3.6]	11 Brinton, 1 Demmler, 26 Hemminki, 15 Hsing, 30 Landgren, 8 Mellemkjaer, 50 Ye
		Small Bowel	19/24.753 (0.08) [1.1]	1 Brinton, 1 Chan, 1 Demmler, 9 Hemminki, 5 Landgren, 2 Mellemkjaer
		Colon	271/30.789 (0.9) [10.3]	1 Armbrecht, 4 Borch, 34 Brinton, 4 Chan, 72 Hemminki, 1 Hoffman, 36 Hsing, 59 Landgren, 50 Mellemkjaer, 1 Mosbech, 9 Talley
		Rectum	121/30.036 (0.4) [4.8]	1 Arvanitakis, 1 Borch, 1 Brinton, 1 Demmler, 35 Hemminki, 25 Hsing, 38 Landgren, 19 Mellemkjaer,
		Anus	3/11.839 (0.02) [4.4]	3 Hemminki
		Liver	40/18.589 (0.2) [4.2]	1 Borch, 2 Brinton, 1 Chan, 14 Hsing, 18 Landgren, 3 Mellemkjaer, 1 Mosbech,

		Biliary tract	30/15.478 (0.2) [6.7]	1 Borch, 4 Brinton, 1Chan, 18 Hsing, 6 Mellemkjaer
		Pancreas	123/33.483 (0.4) [4.8]	5 Borch, 7 Brinton, 3 Demmler, 34 Hsing, 25 Landgren, 19 Mellemkjaer, 30 Shah
		Other	19/5.522 (0.4) [24.9]	17 Borch, 2 Brinton
Female reproductive	126/15.656 (0.8) [11.9]	Breast	79/15.656 (0.5) [7.5]	1 Brinton, 1 Chan, 1 Demmler, 36 Hsing, 39 Mellemkjaer, 1 Mosbech
		Uterus	31/10.495 (0.3) [4.9]	2 Chan, 1 Demmler, 16 Hsing, 11 Mellemkjaer, 1 Mosbech
		Ovary	16/9.827 (0.2) [3.5]	1 Demmler, 9 Hsing, 6 Mellemkjaer
Male reproductive	191/15.189 (1.3) **[31.9]**	Prostate	191/15.189 (1.3) [31.9]	90 Brinton, 3 Hoffman, 58 Hsing, 39 Mellemkjaer, 1 Mosbech
Urinary	101/20.149 (0.5) [8.4]	Kidney	41/14.988 (0.3) [5.2]	10 Brinton, 1 Demmler, 16 Hsing, 14 Mellemkjaer
		Bladder	60/14.988 (0.4) [7.6]	24 Brinton, 1 Demmler, 12 Hsing, 23 Mellemkjaer,
Hematopoietic	199/29.713 (0.7) [10.0]	Hodgkin Lymphoma	3/9.678 (0.03) [2.4]	1 Brinton, 2 Hsing
		Non Hodgkin Lymphoma	70/22.240 (0.3) [15.4]	45 Fallah, 11 Hsing, 14 Mellemmkjaer
		Multiple Myeloma	28/14.888 (0.2) [6.5]	9 Brinton, 1 Hoffman, 11 Hsing, 7 Mellemkjaer,
		Leukemia	76/16.557 (0.5) [10.8]	9 Blackburn, 31 Brinton, 1 Elsborg, 21 Hsing, 13 Mellemkjaer, 1 Siurala
		Other	22/11.808 (0.2) [6.1]	1 Blackburn, 6 Brinton, 2 Chan, 2 Hoffman, 11 Hsing
Skin	86/14.750 (0.6) **[32.8]**	Melanoma	21/14.750 (0.1) [8.0]	12 Brinton, 5 Hsing, 4 Mellemkjaer
		Non melanoma	65/9.589 (0.7) **[61.6]**	8 Hsing, 57 Mellemkjaer
Bone and soft tissue	13/5.766 (0.2) [4.9]	Bone	6/5.161 (0.1) [17.1]	6 Brinton
		Connective/soft tissue	7/5.766 (0.1) [2.6]	5 Brinton, 1 Chan, 1 Demmler

Table 6 - Cancer incidence per organ system in PA patients. Cancer cases not included: 1 kidney/lymphoma/neurinoma (Arvanitakis); 3 genital (not specified if female or male from Brinton); 24 not specified (Borch); 26 not specified (Brinton); 44 not specified (Blackburn); 1 metastatis (Demmler).